

IN THE CLAIMS:

Please amend the following claims 1, 2, 9, 25, 39, 53, 59, 60, 63, and 64 as follows:

1. (Amended) In a method for forming multiple copies of a target sequence of a target polynucleotide, said method comprising the steps of combining a sample with reagents for forming said multiple copies, subjecting said sample to polynucleotide amplification conditions sufficient to form the multiple copies if the target sequence is present in said sample, said reagents comprising an oligonucleotide primer and polymerase; and forming extension products of the oligonucleotide primer at least along said target sequence or along an extended oligonucleotide primer complimentary to said oligonucleotide primer, said extension products being copies of said target sequence, the improvement which comprises forming said extension products in the presence of a second polynucleotide, to which said oligonucleotide primer hybridizes except for a 3'-mismatch on said oligonucleotide primer, under polynucleotide amplification conditions wherein the extension of said oligonucleotide primer along said second polynucleotide is [controlled] suppressed by the 3'-mismatch relative to the extension of said oligonucleotide primer along said target sequence; wherein said target polynucleotide comprises inverted repeat structures.

2. (Amended) In a method for amplifying a target sequence of a target polynucleotide, said method comprising combining a sample suspected of containing said target polynucleotide with reagents for amplifying said target sequence if present and subjecting said combination to polynucleotide amplification conditions wherein said target sequence if present is amplified, said reagents comprising primer A and primer B and a polymerase having 3' to 5' exonuclease activity, the improvement which comprises (a) including in said combination a control polynucleotide, to which said primer A hybridizes except for 1-10 nucleotides at the 3'-end of said primer, [and 3' to 5' exonuclease when said polymerase does not comprise a 3' to 5' exonuclease,] wherein said primer extends along said target sequence and extends along said control polynucleotide to produce copies of said control polynucleotide only after said 1 to 10 nucleotides are degraded by said polymerase having said 3' to 5' exonuclease activity and (b)

*E  
Conclu*  
detecting the presence of said copies of said control polynucleotide, the presence thereof indicating that said reagents and polynucleotide amplification conditions for amplifying said target sequence are functional.

---

9. (Amended) In a method for forming multiple copies of a target sequence of a single stranded target polynucleotide ("target sequence"), said method comprising:

- (a) hybridizing to the 3'-end of said target sequence a first oligonucleotide primer ("first primer"),
- (b) extending, in the presence of a polymerase having 3' to 5' exonuclease activity, said first primer along at least said target sequence, said first primer being capable of hybridizing to, and being extended along, (1) said extended first primer or (2) an extended second oligonucleotide primer ("second primer") wherein said extended second primer results from the extension of a second primer capable of hybridizing to and extending along a polynucleotide that is complementary (complementary polynucleotide) to said target sequence,
- (c) dissociating said extended first primer from said target sequence,
- (d) hybridizing, to the 3'-end of said extended first primer, said first or said second primer,
- (e) extending said first or said second primer along said extended first primer, JUL 08 2000
- (f) dissociating said extended first primer or said extended second primer from said extended first primer,
- (g) hybridizing, to the 3'-end of said extended first or second primer, said first primer, and
- (h) repeating steps (e)-(g), the improvement comprising (i) including, as a positive internal control in the same reaction mixture subjected to steps (a) - (g) above, a control polynucleotide, to which said first or said second primer hybridizes except for 1-10 nucleotides at the 3'-end of said first or said second primer, [and a 3' to 5' exonuclease when said polymerase does not comprise a 3' to 5' exonuclease,] wherein said first or said second primer extends along said control polynucleotide to produce copies of said control polynucleotide only after said 1-10 nucleotides are degraded by said polymerase having 3' to 5' exonuclease activity and (ii) detecting said copies of said control polynucleotide, wherein steps (a)-(h) are performed under polynucleotide amplification conditions.

25. (Amended) A method for forming multiple copies of at least one double stranded polynucleotide ("polynucleotide"), said polynucleotide comprising a single stranded target polynucleotide sequence ("target sequence") and its complementary sequence (complementary sequence), said method having a positive internal control, said method comprising:

(a) treating a sample suspected of containing one or more of said double stranded polynucleotides with (i) at least two oligonucleotide primers capable of hybridizing to a portion of each target sequence and its complementary sequence suspected of being present in said sample under polynucleotide amplification conditions for hybridizing said primers to and extending said primers along said target sequence and said complementary sequences, wherein said primers are selected such that the extension product formed from one primer (primer A), when it is dissociated from its complement, can serve as a template for the formation of the extension product of another primer (primer B), (ii) a control polynucleotide, as a template to which either primer A or B hybridizes except for 1-10 nucleotides of the primer at the 3'-end (control primer), and (iii) a polymerase having 3' to 5' exonuclease activity wherein said primers extend, respectively, along said target sequence and said complementary sequence and the control primer extends along said control polynucleotide only after said 1-10 nucleotides are degraded by said 3' to 5' exonuclease activity,

(b) dissociating primer extension products from their respective templates to produce single stranded molecules and

(c) treating the single stranded molecules produced in step (b) with the primers of step (a) under polynucleotide amplification conditions such that a primer extension product is formed using the single strands produced in step (b) as templates, resulting in amplification of the target sequences and complementary sequences if present, said polynucleotide amplification conditions allowing for the extension of the control primer along said control polynucleotide to provide said positive internal control.

39. (Amended) A method of producing multiple copies of a target sequence of a target polynucleotide, which comprises:

(a) providing in combination (1) a single stranded polynucleotide having a sequence that is said target sequence and that is flanked at each end by at least partially complementary first and

second flanking sequences, (2) an oligonucleotide primer at least a 10 base portion of which at its 3'-end is hybridizable to that member of said first and second flanking sequences that is at the 3'-end of said single stranded polynucleotide, (3) nucleoside triphosphates, (4) a control polynucleotide, as a template to which said oligonucleotide primer hybridizes except for 1-10 nucleotides at the 3-end of said oligonucleotide primer, and (5) a polymerase having 3' to 5' exonuclease activity wherein said primer extends along said target sequence and said primer extends along said control polynucleotide only after said 1-10 nucleotides are degraded by said 3' to 5' exonuclease activity,

(b) incubating said combination under polynucleotide amplification conditions for either wholly or partially sequentially or concomitantly (1) dissociating said single stranded polynucleotide from any complementary sequences, (2) hybridizing said oligonucleotide primer with the flanking sequence at the 3'-end of said single stranded polynucleotide and with said control polynucleotide, (3) extending said oligonucleotide primer along said single stranded polynucleotide to provide a first extended oligonucleotide primer and degrading said oligonucleotide primer hybridized to said control polynucleotide and extending said degraded oligonucleotide along said control polynucleotide, (4) dissociating said first extended primer and said single stranded polynucleotide and dissociating said control polynucleotide and said extended degraded primer, (5) hybridizing said first extended oligonucleotide primer with said oligonucleotide primer and hybridizing said oligonucleotide primer and said control polynucleotide, (6) extending said oligonucleotide primer along said first extended oligonucleotide primer to provide a second extended oligonucleotide primer and degrading said oligonucleotide primer hybridized to said control polynucleotide and extending said oligonucleotide primer along said control polynucleotide to provide an extended degraded primer, (7) dissociating said second extended oligonucleotide primer from said first extended oligonucleotide primer and said extended degraded primer from said control polynucleotide, and (8) repeating steps (5)-(7) above, and (c) detecting the presence of said extended degraded primer, the presence thereof indicating that said reagents and polynucleotide amplification conditions for producing multiple copies of said target sequence of a target polynucleotide are functional.

*H  
E  
Concl'd*

53. (Amended) A kit comprising in packaged combination:

- (a) an oligonucleotide primer,
- (b) a control polynucleotide having a sequence to which said oligonucleotide primer hybridizes except for 1-10 nucleotides at the 3-end of said oligonucleotide primer,
- (c) a modified oligonucleotide primer that is substantially identical to said oligonucleotide primer except for a chemical modification at its 3'-end that prevents degradation, by a 3' to 5' exonuclease, of said 1 to 10 nucleotides,
- (d) nucleoside triphosphates, and
- (e) a polymerase having 3' to 5' exonuclease activity.

59. (Amended) The method of claim 1, wherein the extension is further controlled along said second polynucleotide by contacting the 3'-mismatch with a polymerase having 3' to 5' exonuclease activity.

60. (Amended) The method of claim 59, wherein the extension is further controlled by digesting the 3'-mismatch with the 3' to 5' exonuclease activity and extending the oligonucleotide primer along the second polynucleotide under the polynucleotide amplification conditions.

63. (Amended) The method of claim 1, wherein the target sequence is single-stranded and [comprises] said invert repeat structures that hybridize to the oligonucleotide primer under the polynucleotide amplification conditions.

64. (Amended) The method of claim 1, wherein the target sequence is double-stranded and [comprises] said inverted repeat structures [that] form a stem-loop [and hybridize] that hybridizes to the oligonucleotide primer under the polynucleotide amplification conditions.

#### REMARKS

As an initial matter, Applicant gratefully acknowledges withdrawal of previous rejections under 35 USC §112, first and second paragraphs.